NATURAL TOXINS 5:74–79 (1997)

# Domoic Acid is a Potent Neurotoxin to Neonatal Rats

**Dan Xi,**[1,2] **Yong-Gang Peng,**[1,2] **and John S. Ramsdell**[1,2]*
[1]Marine Biotoxins Program, NOAA Southeast Fisheries Science Center Charleston Laboratory, Charleston, South Carolina
[2]Marine Biomedical and Environmental Sciences, Medical University of South Carolina, Charleston, South Carolina

**ABSTRACT** Domoic acid induces a time-dependent neuroexcitotoxic effect in neonatal rats characterized by hyperactivity, stereotypic scratching, convulsions, and death with observable behaviors occurring at exposures 40 times lower by body weight in neonates than reported in adults. Low doses of domoic acid (0.1 mg/kg) induced c-fos in the central nervous system which was inhibited in part by 2-amino-5-phosphonovaleric acid, an NMDA receptor antagonist. Domoic acid caused no evidence of structural alteration in the brain of neonates as assessed by Nissel staining and cupric silver histochemistry. Domoic acid induced reproducible behavioral effects at doses as low as 0.05 mg/kg and induced seizures doses as low as 0.2 mg/kg. Determination of serum domoic acid levels after 60 min exposure indicated that serum levels of domoic acid in the neonates corresponded closely to the serum levels that induce similar symptoms in adult rats and mice. We conclude that neonatal rats are highly sensitive to the neuroexcitatory and lethal effects of domoic acid and that the increased sensitivity results from higher than expected serum levels of domoic acid. These findings are consistent with other findings that reduced serum clearance of domoic acid is a predisposing factor to domoic acid toxicity. *Nat. Toxins 5:74–79, 1997.* 

**Key Words:** domoic acid; neonate; glutamate receptor; c-fos

## INTRODUCTION

Domoic acid, a rigid analog of glutamate and kainic acid (KA), was responsible for one of the largest human neurotoxic exposures of the past decade [Wright et al., 1989; Perl et al., 1990]. Anterograde memory deficits were the prominent severe neurological symptom [Teitlebaum et al., 1990]. Neuronal damage, particularly in the hippocampus and its associated regions, was found in each of the four postmortem examinations. Although no additional reports of severe human exposures have appeared since 1987, a second outbreak affecting seabirds in Monterey Bay, California, occurred in the fall of 1991; and domoic acid has been widely found to accumulate in shellfish and crustacea in western and eastern coastal waters of North America.

The neurotoxic hazard of domoic acid has been extensively investigated in adult animals, with structural damage being observed in the hippocampus and associated regions in several species [Tryphonas and Iverson, 1990; Tryphonas et al., 1990; Scallet et al., 1993; Peng et al., 1994]. Among the key questions that remain is whether specific age groups are at a greater risk to domoic acid exposure. Perl et al. [1990] observed that the four mortalities were elderly subjects, all over 70 years in age. Overall, the older adults were more likely to suffer memory loss. Pharmacokinetic studies have indicated that domoic acid is rapidly cleared from serum, and that agents that disrupt kidney function increase the toxicity of domoic acid [Truelove and Iverson, 1994; Suzuki and Hierlihy, 1993]. Thus one factor associated with the increased toxicity in older age groups may be decreased renal clearance of domoic acid. Dakshinamurti et al. [1993] have reported neurotoxic effects of domoic acid administered during the middle embryonic period of mice. No neurotoxic effects of domoic acid were evident at birth in this study, but severe reorganization of the hippocampus was evident by day 21 in the mice exposed *in utero*. The neonatal period represents an age group that is differentially susceptible to many neurotoxins. During this period the blood brain barrier is incomplete. Domoic acid is poorly permeable into brain tissue in the adult animals [Preston and Hynie, 1991] and, thus, there is reason to expect greater toxicity of domoic acid to neonates. On the other hand, neonates tend to be more resistant to the effects of certain neurotoxins that induce neuronal degeneration [Strafstrom et al., 1992]; and, for this reason, they may be less susceptible to certain sustained neurotoxic effects than adults.

Contract grant sponsor: NIH; Contract grant number: DK 43107; Contract grant sponsor: National Oceanographic and Atmospheric Administration.

*Correspondence to: John S. Ramsdell, Ph.D., Marine Biomedical and Environmental Sciences, Medical University of South Carolina, 221 Fort Johnson Road, Charleston, SC 29412.

Received 3 March 1996; Accepted 23 January 1997

In this study we have investigated the neurotoxic effects of domoic acid on neonatal rats. We conducted dose response studies using the $LD_{50}$ and a observable behavioral toxicity score as endpoints. We next evaluated the basis for the high susceptibility of neonates to domoic acid by examining serum levels of domoic acid. We determined that neonates accumulated high circulating levels of domoic acid and this paralleled their high susceptibility to the toxin.

## EXPERIMENTAL PROCEDURES

### Experimental Animals

Long-Evans lactating rats with newborn pups (Charles River Lab.) were housed with food and water available ad libitum, and were maintained on a 12 hr light-dark cycle. The pups received a single intraperitoneal (i.p.) injection of domoic acid (Sigma Chemical Co., St Louis, MO) in saline. Controls were administered as an equal amount of saline vehicle only.

### c-fos Northern Analysis

Rats were anesthetized with sodium pentobarbital at 60 min following the i.p. injection of domoic acid. The whole brain was rapidly dissected and immediately frozen in liquid nitrogen and stored at −80°C for RNA extraction. Total RNA was prepared with TRI-reagent, a single-step RNA isolation. Thirty micrograms of total RNA of each sample was loaded in a 1% agarose formaldehyde gel and analyzed by constant current electrophoresis at 60 V. The RNA was then transferred directly and cross-linked onto a nylon membrane. The filter was hybridized at 50°C with random labeled rat c-fos probe overnight. The membrane was washed at room temperature, and then analyzed by autoradiography with intensifier screens on XAR-5 film. The same filter was rehybridized with mouse β-actin probe, which was used to verify that each lane had the same amount of total RNA loaded.

### Histochemistry for Structural Damage

PND2 rats were anesthetized with sodium pentobarbital at 72 hr following the i.p. injection of domoic acid and intracardially perfused with 4% paraformaldehyde in 0.067 M Na cacodylate buffer (pH 7.2). Brains were removed and embedded in gelatin. Frozen sections (50 μm) were cut and every sixth section stained by one of two procedures. The first procedure stained the sections with thionine to reveal Nissel substance. The second procedure stained the sections using the cupric silver method [de Olmos et al., 1981] as previously described by Peng et al. [1994].

### Serum Level of Domoic Acid Using Radioreceptor Assay

Rats were anesthetized with sodium pentobarbital (40 mg/kg i.p.) at 1 or 2 hr following administration of vehicle or domoic acid, and then blood was collected by cardiac puncture. Domoic acid levels were determined by heterologous radioreceptor assay as previously described [Van Dolah et al., 1997]. Assays were carried out in 96-well polystyrene plates in 50 mM Tris-HCl, pH 7.4, containing 0.1 mg/ml GLURG membranes, 5 nM [$^3$H]-kainic acid (New England Nuclear), and the serum sample in a total volume of 210 μl. Standard calibration solutions (National Research Council, Canada) contained 10 pM to 1 μM domoic acid. Following incubation of samples at 4°C for 1 hr, all wells were filtered onto a 96-grid glass fiber filter mat using a 96-place filtration manifold (Milliblot, Millipore), and rinsed four times with ice-cold Tris buffer (50 mM, pH 7.4). The filter mat was dried on a slide warmer (60°C) for 15 min, impregnated with solid scintillant, and after cooling, was counted directly in a Microbeta 1450 scintillation counter (Wallac, Gaithersburg, MD). The standard curve was derived by a four parameter logistic fit of standard curve and unknown sample concentrations were calculated from the linear portion of this fit.

### Behavioral Rating Scale for Domoic Acid Toxicity

Each rat was placed in a bedding cage (14 × 20 × 26 cm). Rat behavioral changes were recorded following administration of either vehicle or domoic acid using the rating scale of Tasker et al. [1991]. Seven domoic acid symptoms were monitored as described in Peng and Ramsdell [1996] for 60 min by the same observer under blind conditions and the time of onset and duration of each symptom recorded. Each behavior was assigned a score shown in parenthesis: hypoactivity (1), sedation (2), hyperactivity (3), scratching (4), loss of balance control (5), tremors-convulsions (6), and death (7). A toxicity score was calculated as the sum of each behavioral change quantified by the product of the duration (min) of the symptom and the assigned score.

## RESULTS

### Dose Dependency for Domoic Acid Induced Lethality

Neonatal rats were treated with increasing doses of domoic acid i.p. and observed for up to 4 hr. Figure 1 shows the dose-lethality relationship for postnatal day 2 (PND2) and PND10 rats. The $LD_{50}$ for PND2 rats is about 0.25 mg/kg. PND2 rats were more sensitive than PND10 ($LD_{50}$ about 0.7 mg/kg).

### Domoic Acid Induces Neurotoxic Effects in Neonatal Rats

We first determined whether these low doses of domoic acid reached biologically effective levels in the central nervous system using c-fos as a biochemical marker for early biological effect. Domoic acid administered at the sublethal dose of 0.1 mg/kg to PND5 rats caused an increased accumulation of total brain c-fos mRNA (Fig. 2, left lanes). Pretreatment of the animals with the NMDA glutamate subtype antagonist, AP-5, reduced the c-fos mRNA accumulation (Fig. 2, right lanes). These results indicate that domoic acid induces brain c-fos, as has been observed in adult rodents and does so by a pathway that requires NMDA receptors for a maximal effect.

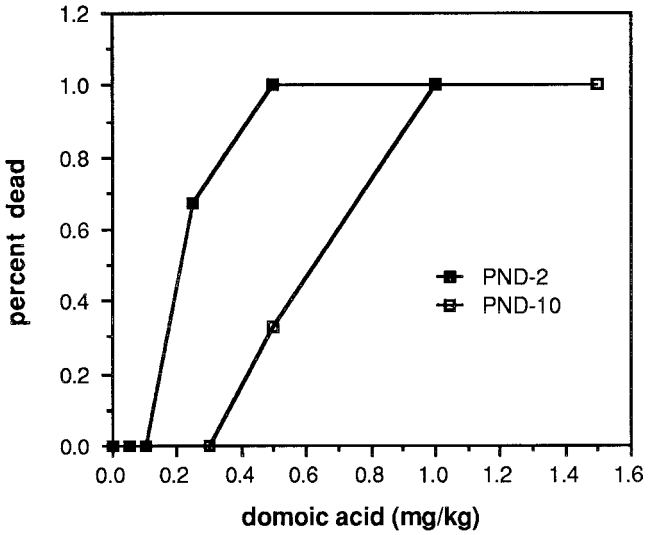


**Fig. 1.** Dose dependency for domoic acid induced lethality in neonatal rats. PND2 and PND10 rats were treated with increasing doses of domoic acid intraperitoneal and observed for up to 4 hr. Data shown are for three independent experiments using a total of 20 PND2 rats and 15 PND10 rats.

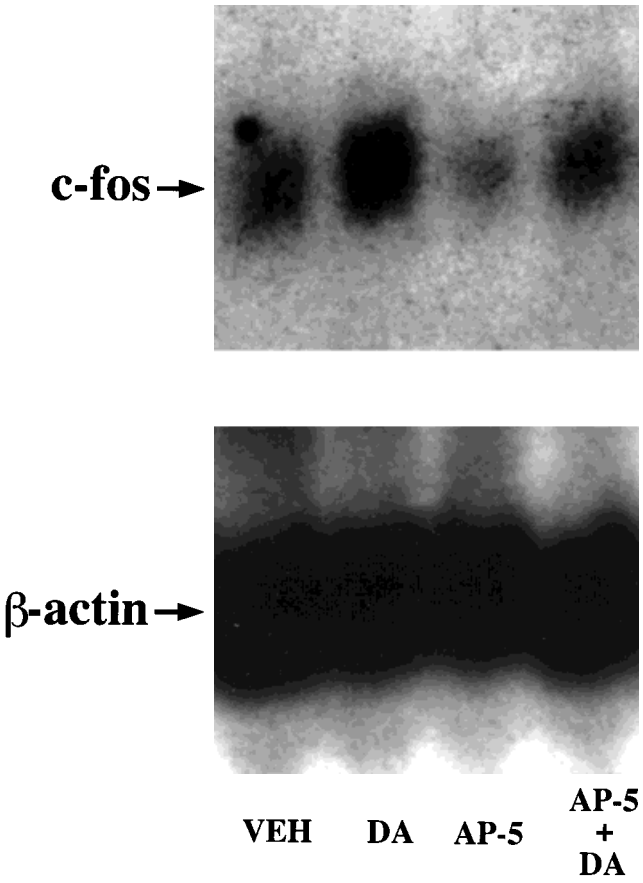


**Fig. 2.** Domoic acid induced c-fos mRNA in the whole brain of PND5 rats. PND5 rats were pretreated with saline vehicle or 1.0 mg/kg AP-5 intraperitoneal for 30 min and then 0.1 mg/kg domoic acid for an additional 90 min. mRNA extracted from whole brain was probed with a full length rat c-fos cDNA random probe (**top**) and with β-actin cDNA (**bottom**). This result shows that AP-5 markedly reduced the induction of c-fos mRNA by domoic acid in the brain. This experiment was repeated three times with similar results.

We next examined the dose and time requirements for typical observable behaviors in response to domoic acid. PND5 rats became hyperactive within 5 min of domoic acid i.p. administration. Stereotypic behavior in the form of scratching occurred within 20 min at a dose of 0.2 mg/kg, twice as long at 0.1 mg/kg, and did not occur at 0.02 mg/kg. Tonic/clonic seizures began around 40 min at 0.2 mg/kg. These results indicate that domoic acid induces similar symptomatic changes in neonate rats as is found in adults, but at doses substantially lower than reported in adult rats.

We next assessed structural neurological damage following domoic acid exposure. PND2 rats were treated with 0.2 mg/kg (LD50 = 0.25 mg/kg) and five surviving animals were examined after 72 hr. Nissl staining revealed no evidence of morphological changes in the hippocampus of domoic acid treated animals. Analysis of serial sections failed to reveal evidence of structural damage by cupric silver impregnation histochemistry (data not shown). Both of these methods show neurological damage in rodents and primates within 72 hr of sublethal domoic acid exposure.

### Dose Dependency of Domoic Acid Effect on Behavioral Symptomatology

Since neonatal rats exhibited behavioral symptoms to domoic acid comparable to those observed in juvenile and adult rats and mice, we quantified the sublethal toxicity using the behavioral toxicity scoring method developed for mice by Tasker et al. [1991]. Three litters of neonatal rats were treated with four sublethal doses of domoic acid. Half of each litter was treated on PND5 and the other half treated on PND10. Domoic acid caused a concentration-dependent increase in toxicity score, with the lowest observable effect at 0.05 mg/kg (Fig. 3). No reproducible differences were observed in the sensitivity or maximal response between PND5 and PND10 rats. We next measured domoic acid in serum collected from these animals after the 60 min observation period.

### Serum Levels of Domoic Acid in Neonatal Rats

Serum was collected at 60 min post-treatment following the conclusion of behavioral symptomatology study and quantified by the domoic acid receptor assay. Serum levels increased in a concentration-dependent manner (Fig. 4). No reproducible differences were observed in the sensitivity or maximal response between PND5 and PND10 rats. We next established the relationship between serum levels and toxicity score. Observable toxicity increased as a function of serum domoic acid (Fig. 5). Using serum and toxicity data from adult mice [Peng and Ramsdell, 1996], the data from the adult mice overlapped that of the neonatal rat. This

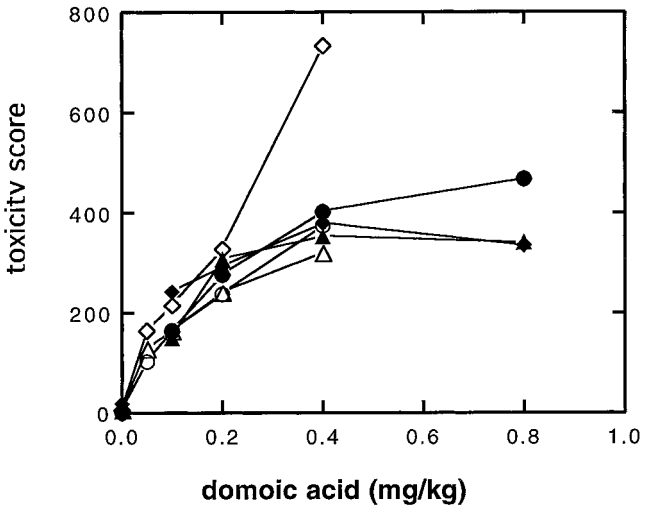


**Fig. 3.** Behavioral changes in neonatal rats in response to different external dose domoic acid exposures. PND5 (open symbols) or PND10 (closed symbols) were treated (i.p.) with increasing (0.05 to 0.4 mg/kg) of domoic acid or saline vehicle and then sacrificed at 60 min. Behavioral changes were monitored continuously for the 60 min period under blind conditions and a toxicity score calculated. Results are the mean serum concentrations of domoic acid from three independent experiments (circles, diamonds, and triangles) using one PND5 and one PND10 littermate rat per dose for each experiment.

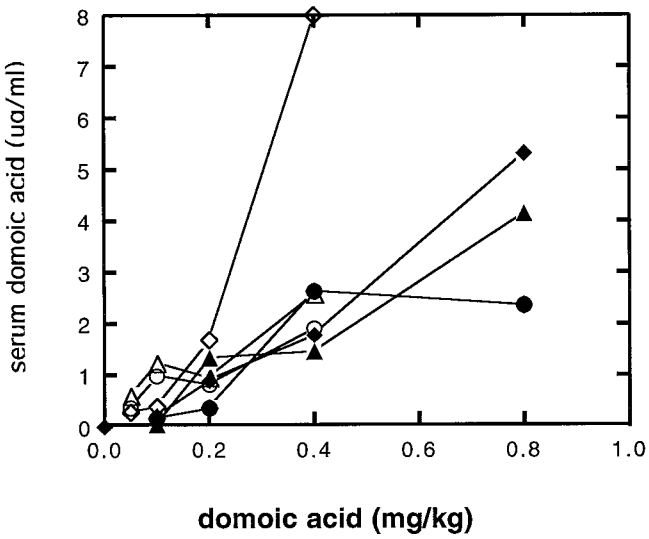


**Fig. 4.** Serum levels of domoic acid in neonatal rats in response to different external dose domoic acid exposures. PND5 (open symbols) or PND10 (closed symbols) were treated (i.p.) with increasing (0.05 to 0.4 mg/kg) of domoic acid or saline vehicle and then sacrificed at 60 min. Domoic acid was measured by radioreceptor assay. Results are the mean serum concentrations of domoic acid from three independent experiments (circles, diamonds, and triangles) using one PND5 and one PND10 littermate rat per dose for each experiment.

indicates that the neonates and adults have similar sensitivity to internal levels of domoic acid and that the higher susceptibility of neonates results from retention of higher levels of the toxin. Taken together these results support the hypothesis that reduced clearance of domoic acid is a predisposing factor to the increased sensitivity of neonatal rats to domoic acid.

## DISCUSSION

This study provides the first investigation of domoic acid action in neonatal animals. Previous studies have been conducted during the embryonic and midgestational periods. Our results demonstrate that neonates respond to domoic acid with characteristic symptoms, biochemical indicators, and pharmacological specificity. However, the neonates are more sensitive to domoic acid per body weight than are adults. The relationship between internal dose and effect is consistent between the two age groups, but the relationship between internal dose and external dose is shifted in the neonate. Our results support the hypothesis that reduced serum clearance is a contributing factor in the sensitivity of neonates to domoic acid toxicity.

Domoic acid (i.p.) induces behavioral changes and neuroexcitotoxic effects on hippocampal pyramidal cells [Tryphonas et al., 1990; Sutherland et al., 1990]. We find that neonatal rats develop comparable symptoms as observed in adult rats, but are substantially more sensitive to domoic acid per body weight. Based on the data of Tryphonas et al. [1980] for adult rats and the data of this report for neonatal rats, neonates are approximately 80-fold more sensitive to domoic acid induced scratching and approximately 40-fold more sensitive to domoic acid induced seizures. By contrast, teratogenic and embryonic studies have not indicated a substantially greater lethality of these developmental periods beyond that of maternal toxicity [Khera et al., 1994; Dakshinamurti et al., 1993]. Using kainic acid exposure as a model for domoic acid toxicity, old and middle-aged rats were found to be four- and twofold more sensitive, respectively, than young juveniles [Wozniak et al., 1991]. A higher sensitivity of neonates to domoic acid is similar to the kainic acid effect on developing rats which has been proposed to result from the greater bioavailability of kainic acid [Stafstrom et al., 1992].

Domoic acid induces brain c-fos in animals at doses comparable to the early observed symptomatic effects [Peng and Ramsdell, 1996]. This indicates that the low dose effects of domoic acid can result from its effects on the central nervous system. The results from this report support a comparable situation in neonates. The mechanism by which neonates have greater sensitivity to domoic acid effects may be at the level of the sensitivity of the target cell or be the concentration that the toxin achieves at the target cell. Hippocampal pyramidal cells are a primary *in vivo* target for domoic acid [Peng et al., 1994]. Hippocampal cells isolated from PND2 rats are sensitive to concentrations of domoic acid as low as 50 nM [Xi and Ramsdell, 1996]. Based upon the radioligand data of Hampson et al., [1992] with rat forebrain membranes, 50 nM domoic acid would occupy

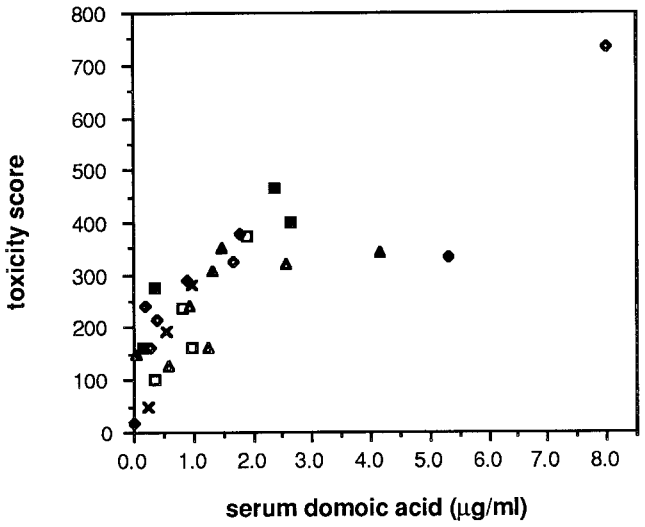


**Fig. 5.** Behavioral changes in neonatal rats in response to different internal dose domoic acid exposures. The toxicity score and serum domoic acid level data for PND5 (open symbols) or PND10 (closed symbols), and adult mice (crosses) were plotted independent of external exposure. Data from the neonatal rats is the same set used for Figures 3 and 4 and the data from the adult mice is from Peng and Ramsdell (1996).

80–90% of kainic selective glutamate receptors. Based upon the pharmacokinetic study of Preston and Hynie [1991], 50 nM domoic acid is close to the calculated brain level of domoic acid (154–245 nM) following a near threshold intravenous dose of 1 mg/kg in adult rats. This indicates that the neonatal target cells cannot fully account for the higher sensitivity of neonates to domoic acid. Thus the sensitivity of neonates to domoic acid more likely results from greater bioavailability of domoic acid in neonates.

Increased bioavailability of domoic acid in neonates could result from an incomplete blood brain barrier. The charged structure of domoic acid would predict its having poor permeability to the central nervous system, unless it entered by a transport mechanism. Preston and Hynie [1991] have examined the transfer of radiolabeled domoic acid into the brain. They determined that domoic acid will enter the central nervous system from the blood, but does so with a high transfer constant, exceeding that of sucrose. This indicates that domoic acid does not enter the brain by a selective uptake mechanism. The high sensitivity of neonates to domoic acid is similar to that reported for kainic acid [Stafstrom et al., 1992]. This study suggested that the sensitivity of the neonates resulted from greater access of the toxin through the underdeveloped blood-brain barrier based upon the observation that surgical disruption of the blood-brain barrier did not decrease the latency of appearance of symptoms. However, the results presented here with neonates taken together with existing data in adult rodents indicate that the serum levels of domoic acid needed to induce toxic responses is similar in neonates and adults. This suggests that increased accessibility of domoic acid to the brain in neonates is not the major contributing factor to domoic acid toxicity in neonates. Truelove and Iverson [1994] calculated the serum levels of domoic acid necessary to induce scratching after 50 min i.p. exposure of adult rats to be approximately 0.4 to 1.2 μg/ml. In neonates, animals that exhibited scratching after 51 and 40 min had domoic acid serum levels of 0.60 and 0.36 μg/ml, respectively, after 60 min of exposure. Likewise, data from adult mice overlap the neonatal rat data in this report when serum levels are plotted against toxicity score. Another factor that influences the brain uptake of domoic acid is renal clearance [Preston and Hynie, 1991].

Renal clearance of domoic acid has been predicted to be a major predisposing factor to domoic acid toxicity. Domoic acid is cleared rapidly from the serum by nearly 90% within 60 min in rats [Preston and Hynie, 1991; Truelove and Iverson, 1994]. Disrupting renal clearance of domoic acid increases toxicity and both surgical and pharmacological disruption of kidney clearance increases the circulating levels of domoic acid [Preston and Hynie, 1991; Robertson, et al., 1992; Suzuki and Hierlihy, 1993]. We find that neonatal rats exhibit higher levels of serum domoic acid after 60 min i.p exposure, than we would anticipate based on existing data for adult mice [Peng and Ramsdell, 1996]. In adult mice, serum levels of at least 1 μg/ml domoic acid are maintained for up to 60 min with a 4 mg/kg exposure. By contrast 1 μg/ml serum concentration is maintained for 60 min at doses as low as 0.1 mg/kg (in 2 of 6 animals) and in all neonatal rats given 0.4 mg/kg. These results indicate that neonates maintain greater serum levels for domoic acid than do adults for a given dose, and that the increased serum levels of domoic acid in neonates accounts at least in part for their greater sensitivity.

Our results identify neonatal rats as forty times more sensitive to the neuroexcitatory and lethal effects of domoic acid than adults. Serum clearance appears to be the major predisposing factor to the high susceptibility of neonates to domoic acid toxicity. Domoic acid has been demonstrated to accumulate in the urine with a half maximal time between 90 and 360 min; however, accumulation in other fluid components has not been fully investigated. Exposure of neonates to domoic acid by suckling represents a potential route of exposure to that requires consideration given the much greater sensitivity of neonates.

## ACKNOWLEDGMENTS

We thank Elizabeth Fairey for assisting with toxicity scoring and Dr. David Kurtz for providing the rat c-fos probe. This work was supported by funds provided by the National Institutes of Health (DK 43107) and the National Oceanographic and Atmospheric Administration.

## REFERENCES


Dakshinamurti K, Sharma SK, Sundaram M, Watanabe T (1993): Hippocampal changes in developing postnatal mice following intrauterine exposure to domoic acid. J Neurosci 13:4486–4495.

de Olmos JS, Ebbesson SOE, Heimer L (1981): Silver methods for the impregnation of degenerating axoplasm. In Heimer L, Robards M (eds): Neuroanatomical Tract Tracing Methods. New York: Plenum Press, pp 117–120.

Hampson DR, Huang XP, Wells JW, Walter JA, Wright JLC (1992): Interaction of domoic acid and several derivatives with kainic acid and AMPA binding sites in rat brain. Eur J Pharmacol 218:1–8.

Khera, KS, Whalen C, Angers G, Arnold DL (1994): Domoic acid: A teratology and homeostatic study in rats. Bull Environ Contamin Toxicol 53:18–24.

Peng YG, Taylor TB, Finch RE, Switzer RC, Ramsdell JS (1994): Neuroexcitatory and neurotoxic actions of the amnesic shellfish poison, domoic acid. NeuroReport 5:981–985.

Peng YG,, Ramsdell JS (1996): Brain Fos is a sensitive biomarker for the lowest observed neuroexcitatory effects of domoic acid. Fundam Appl Toxicol 31:162–168.

Perl TM, Bédard L, Kosatsky T, Hockin JC, Todd ECD, Remis RS (1990): An outbreak of toxic encephalopathy caused by eating mussels contaminated with domoic acid. N Engl J Med 322:1775–1780.

Preston E, Hynie I (1991): Transfer constants for blood-brain barrier permeation of the neuroexcitatory shellfish toxin, domoic acid. Can J Neurol Sci 18:39–44.

Robertson H, Renton K, Kohn J, White T (1992): Patterns of Fos expression suggest similar mechanisms of action for the excitotoxins domoic acid and kainic acid. Ann NY Acad Sci 648:330–334.

Scallet AC, Binienda Z, Caputo FA, Hall S, Paule MG, Roundtree RL, Schmued L, Sobotka L, Slikker W (1993): Domoic acid-treated cynomolgus monkeys (M. fascicularis): Effects of dose on hippocampal neuronal and terminal degeneration. Brain Res 627:307–313.

Stafstrom CE, Thompson JL, Holmes GL (1992): Kainic acid seizures in the developing brain: Status epilepticus and spontaneous recurrent seizures. Dev Brain Res 65:227–236.

Sutherland RJ, Hoesing JM, Whishaw IQ (1990): Domoic acid, an environmental toxin, produces hippocampal damage and severe memory impairment. Neurosci Lett 120:221–223.

Suzuki CAM, Hierlihy SL (1993): Renal clearance of domoic acid in the rat. Food Chem Toxicol 701–706.

Tasker RAR, Connell BJ, Strain SM (1991): Pharmacology of systemically administered domoic acid in mice. Can J Physiol Pharmacol 69:378–382.

Teitelbaum JS, Zatorre RJ, Carpenter S, Gendron D, Evans AC, Gjedde A, Cashman NR (1990): Neurologic sequelae of domoic acid intoxication due to the ingestion of contaminated mussels. N Engl J Med 322:1781–1787.

Truelove J, Iverson F (1994): Serum domoic acid clearance and clinical observations in the cynomolgus monkey and Sprague-Dawley rat following a single IV dose. Bull Environ Contam Toxicol 52:479–486.

Tryphonas L, Iverson F (1990): Neuropathology of excitatory neurotoxins: The domoic acid model. Toxicol Pathol 18:165–169.

Tryphonas L, Truelove J, Nera E, Iverson F (1990): Acute neurotoxicity of domoic acid in the rat. Toxicol Pathol 18:1–9.

Van Dolah FM, Leighfield TA, Haynes BL, Hampson DR, Ramsdell JS (1997): A microplate receptor assay for amnesic shellfish poisoning toxin, domoic acid, utilizing a cloned glutamate receptor. Analytical Biochem 245:102–105.

Wozniak D, Stewart GR, Miller JP, Olney JW (1991): Age-related sensitivity to kainate neurotoxicity. Exper Neurol 114:250–253.

Wright JLC, Boyd RK, deFreitas ASW, Falk M, Foxall RA, Jamieson WD, Laycock MV, McCulloch AW, McInnes AG, Odense P, Pathak VP, Quilliam MA, Ragan MA, Sim PG, Thibault P, Walter JA, Gilgan M, Richard DJA,, Dewar D (1989): Identification of domoic acid, a neuroexcitatory amino acid, in toxic mussels from eastern Prince Edward Island. Can J Chem 67:481–490.

Xi D, Ramsdell JS (1996): Glutamate receptors and calcium entry mechanisms for domoic acid in hippocampal neurons. NeuroReport 7:1115–1120.